5

10

15

30

CLAIMS:

- 1. A method of separating an antibody from a mixture of antibody and at least one contaminant, the method comprising:
- (a) placing the antibody and contaminant mixture in a first solvent stream, the first solvent stream being separated from a second solvent stream by an electrophoretic membrane having a molecular mass cut-off of 50 to 150 kDa;
- (b) selecting a pH of 7.5 to 9.5 for the first solvent stream such that contaminants with an isoelectric point (pI) lower than the antibody to be separated will be charged;
- (c) applying an electric potential between the two solvent streams causing movement of at least some of the contaminants through the membrane into the second solvent stream while the antibody is substantially retained in the first solvent stream, or if entering the membrane, being substantially prevented from entering the second solvent stream;
 - (d) optionally, periodically stopping and reversing the electric potential to cause movement of any antibody having entered the membrane to move back into the first solvent stream, wherein substantially not causing any contaminants that have entered the second solvent stream to re-enter the first solvent stream; and
- (e) repeating step (d) and optionally step (d) until the first solvent stream contains the desired purity of antibody, wherein the recovery of the antibody is at least 70% and the antibody being less denatured or altered compared with the same antibody obtained by conventional antibody purification methods.
- 25 2. The method according to claim 1 wherein the antibody and contaminant mixture is a monoclonal antibody in ascitic fluid.
 - 3. The method according to claim 1 or 2 wherein the electrophoretic membrane has a molecular mass cut-off of 100 kDa.
 - 4. The method according to any one of claims 1 to 3 further including the steps of:
 - (f) placing the separated antibody in a fresh first solvent stream, the fresh first solvent stream being separated from a second solvent stream by an electrophoretic membrane having a molecular mass cut-off larger than that of the electrophoretic membrane used in step (b);
- 35 (g) selecting a pH of the fresh first solvent stream such that the pH is within 1 pH unit of the pI of the antibody;



5

10

15

20

30

35

- (h) applying an electric potential between the two solvent streams causing movement of at least some of the contaminants through the membrane into the second solvent stream while the antibody is substantially retained in the fresh first solvent stream, or if entering the membrane, being substantially prevented from entering the second solvent stream;
- (i) optionally, periodically stopping and reversing the electric potential to cause movement of any antibody having entered the membrane to move back into the fresh first solvent stream, wherein substantially not causing any contaminants that have entered the second solvent stream to re-enter the fresh first solvent stream; and
- (j) repeating step (h) and optionally step (i) until the fresh first solvent stream contains the desired purity of antibody, wherein the recovery of the antibody is at least 70%.
- 5. The method according to claim 4 wherein the molecular mass cut-off of the electrophoretic membrane used in step (f) is at least 200 kDa.
- 6. The method according to claim 4 wherein the molecular mass cut-off of the electrophoretic membrane used in step (f) is 1000 kDa.
- 7. The method according to any one of claims 4 to 6 wherein the pH of the fresh first solvent stream in step (g) is from 6 to 8.
- 8. The method according any one of claims 4 to 6 wherein the pH of the fresh first solvent stream in step (g) is within 0.5 pH units of the pI of the antibody.
- 9. The method according to any one of claims 1 to 8 wherein recovery of the antibody is at least 90%.
- 25 10. A method of separating an antibody from a mixture of antibody and at least one contaminant, the method comprising:
 - (a) placing the antibody and contaminant mixture in a first solvent stream, the first solvent stream being separated from a second solvent stream by an electrophoretic membrane having a molecular mass cut-off of at least 200 kDa:
 - (b) selecting a pH of the first solvent stream such that the pH is within 1 pH unit of the pI of the antibody;
 - (c) applying an electric potential between the two solvent streams causing movement of at least some of the contaminants through the membrane into the second solvent stream while the antibody is substantially retained in the

5

10

15

20

25

first solvent stream, or if entering the membrane, being substantially prevented from entering the second solvent stream;

- (d) optionally, periodically stopping and reversing the electric potential to cause movement of any antibody having entered the membrane to move back into the first solvent stream, wherein substantially not causing any contaminants that have entered the second solvent stream to re-enter the first solvent stream; and
- (e) repeating step (c) and optionally step (d) until the first solvent stream contains the desired purity of antibody, wherein the recovery of the antibody is at least 70% and the antibody being less denatured or altered compared with the same antibody obtained by conventional antibody purification methods.
- 11. The method according to claim 10 wherein the antibody and contaminant mixture is a monoclonal antibody in ascitic fluid.
- 12. The method according to claim 10 or 11 wherein the molecular mass cut-off of the electrophoretic membrane used in step (a) is 1000 kDa.
- 13. The method according to any one of claims 10 to 12 wherein the pH of the first solvent stream in step (b) is from 6 to 8.
- 14. The method according to any one of claims 10 to 12 wherein the pH of the first solvent stream is step (b) is within 0.5 pH units of the pI of the antibody.
- 15. The method according to any one of claims 10 to 14 wherein recovery of the antibody is at least 90%.
- 16. An isolated antibody purified by the method according to any one of claims 1 to 15.
- 17. The isolated antibody according to claim 16 being a monoclonal antibody.

